

Johnsson et al.

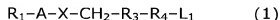
Serial No.: 10/591,162

Date Filed: October 3, 2006

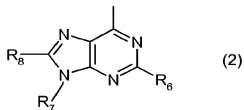
Please amend claims 1, 2 and 11.

In the claims

1. (currently amended) A compound of formula (1)



wherein

the group  $R_1-A$  is a purine radical of formula (2)

X is oxygen;

 $R_1$  is a group  $-R_2-L_2$ ;

$R_2$  and  $R_4$  are, independently of each other, a straight or branched chain alkylene group or polyvalent branched chain alkyl group with 1 to 300 carbon atoms, optionally substituted by a lower alkyl, lower alkoxy, lower acyloxy or halogen wherein optionally

(a) one or more carbon atoms are replaced by oxygen;

(b) one or more carbon atoms are replaced by nitrogen

carrying a hydrogen atom, and the adjacent carbon atom is substituted by oxo;

(c) one or more carbon atoms are replaced by oxygen, and the adjacent carbon atom is substituted by oxo;

(d) the bond between two adjacent carbon atoms is a double or a triple bond;

(e) one or more carbon atoms are replaced by a phenylene, a saturated or unsaturated cycloalkylene, a saturated or unsaturated bicycloalkylene, a divalent heteroaromatic or a divalent saturated or unsaturated heterocyclyl group;

(f) two adjacent carbon atoms are replaced by a disulfide linkage;

or a combination of two or more alkylene and/or modified alkylene groups as defined under (a) to (f) hereinbefore;

R<sub>3</sub> is an aromatic or a heteroaromatic group, or an optionally substituted 1-~~alkylene~~ ~~alkenylene~~, 1-~~alkenylene~~ ~~alkynylene~~, 1-cycloalkenylene, or an unsaturated heterocyclyl group with the double bond connected to CH<sub>2</sub>;

R<sub>6</sub> is hydrogen, hydroxy or unsubstituted amino; one of R<sub>7</sub> or R<sub>8</sub> is R<sub>1</sub> and the other one is hydrogen; and

L<sub>1</sub> and L<sub>2</sub> are the same or different labels and each is selected from the group consisting of a fluorophore or a chromophore, a magnetic probe, a contrast reagent, a radioactive moiety, avidin, streptavidin, biotin, a moiety which is capable of crosslinking to other molecules selected from a maleimide, an activated carboxy group, an azide and a benzophenone; a tethered metal-chelate which is capable of generating hydroxyl radicals upon exposure to H<sub>2</sub>O<sub>2</sub>, ascorbate, malachite green, a solid support, a lipid, methotrexate, a linear poly(arginine) of D- and/or L-arginine with 6-15 arginine residues, oligomers of 6-50 subunits wherein at least one subunit has an attached guanidine group or a peptide having an RKKRRQRRR amino acid sequence (SEQ ID NO: 1); or

L<sub>1</sub> is a bond connecting R<sub>4</sub> to A forming a cyclic substrate; a further group -R<sub>3</sub>-CH<sub>2</sub>-X-A-R<sub>1</sub>; or a nucleic acid or a derivative thereof capable of undergoing base-pairing with its complementary strand; or

if R<sub>7</sub> is a hydrogen then L<sub>2</sub> is capable of undergoing base-pairing with its complementary strand where L<sub>2</sub> is a nucleic acid ~~or a peptide-nucleic acid~~.

2. (currently amended) The compound according to claim 1, wherein R<sub>3</sub> is phenylene, an unsubstituted or substituted mono- or bicyclic divalent heteroaryl group of 5 or 6 rings atoms comprising zero, one, two, three or four ring nitrogen atoms and zero or one oxygen atom and zero or one sulfur atom, with the proviso that at least one ring carbon atom is replaced by a nitrogen, oxygen or sulfur atom, ~~1-alkylene alkenylene, 1-alkenylene alkynylene, 1-cyclohexenylene~~ with 3 to 7 carbon atoms, wherein the double or triple bond is connected to CH<sub>2</sub>, or an optionally substituted unsaturated divalent heterocyclyl group with 3 to 12 atoms and 1 to 5 heteroatoms selected from nitrogen, oxygen and sulfur, and a double bond in the position connecting the heterocyclyl group to CH<sub>2</sub>.

3. (cancelled)

4. (previously presented) The compound according to claim 1, wherein R<sub>3</sub> is phenylene.

5. (previously presented) The compound according to claim 1, wherein R<sub>3</sub> is thienylene.

6. (previously presented) The compound according to claim 1, wherein R<sub>6</sub> is unsubstituted amino, R<sub>7</sub> is R<sub>1</sub>, and R<sub>8</sub> is hydrogen.

7. (cancelled)

8. (previously presented) The compound according to claim 7, wherein  $L_2$  is a fluorophore or a chromophore.
9. (previously presented) The compound according to claim 7, wherein  $L_1$  is a fluorophore or a chromophore and  $L_2$  is a fluorophore or a chromophore.
10. (previously presented) The compound according to claim 9, wherein  $L_1$  is a fluorescence donor and  $L_2$  is a fluorescence quencher or  $L_1$  is a fluorescence quencher and  $L_2$  is a fluorescence donor.
11. (currently amended) The compound according to claim 9 ~~10~~, wherein  $L_1$  and  $L_2$  constitute a FRET pair.
- 12-13. (cancelled)
14. (previously presented) The compound according to claim 1, wherein  $R_6$  is unsubstituted amino,  $R_7$  is hydrogen, and  $R_8$  is  $R_1$ .
15. (previously presented) The compound according to claim 1, wherein  $R_6$  is unsubstituted amino,  $R_7$  is hydrogen, and  $R_8$  is a group  $-R_2-L_2$ .
16. (previously presented) The compound according to claim 15, wherein  $L_2$  is a fluorophore or a chromophore.
17. (previously presented) The compound according to claim 16, wherein  $L_1$  is a fluorophore or a chromophore.

18. (previously presented) The compound according to claim 17, wherein L<sub>1</sub> and L<sub>2</sub> constitute a fluorescence donor or a fluorescence quencher.

19. (previously presented) The compound according to claim 18, wherein L<sub>1</sub> and L<sub>2</sub> constitute a donor or an acceptor in a FRET pair.

20. (previously presented) The compound according to claim 15, wherein L<sub>2</sub> is avidin, streptavidin or biotin.

21. (previously presented) The compound according to claim 15, wherein L<sub>2</sub> is a moiety covalently attached to a solid support.

22. (previously presented) The compound according to claim 15, wherein L<sub>2</sub> is a linear poly(arginine) of D- and/or L-arginine with 6-15 arginine residues, an oligomer of 6-50 subunits wherein at least one subunit has an attached guanidine group or a peptide having an RKKRRQRRR amino acid sequence (SEQ ID NO: 1).

23-43 (cancelled)

44. (previously presented) A method for detecting a protein of interest, wherein the protein of interest is fused to a mutant of a human AGT, the method comprising:

- (a) contacting the AGT fusion protein with a compound of formula (1) according to claim 1; and
- (b) detecting the AGT fusion protein using label L<sub>1</sub> and/or L<sub>2</sub> in a system designed for recognizing and/or handling the label.

45. (previously presented) The method according to claim 44, wherein in the compound of formula (1)  $L_2$  is a solid support, and the AGT fusion protein contacted with the compound of formula (1) is separated from the compound of formula (1) by filtration or centrifugation or separation of magnetic beads.

46. (previously presented) The method according to claim 44, wherein in the compound of formula (1)  $L_1$  is one member and  $L_2$  the other member of two interacting chromophores or fluorophores, wherein energy can be transferred non-radiatively through dynamic or static quenching, and the AGT fusion protein is detected by fluorescence.

47. (previously presented) The method according to claim 44 for detecting a protein of interest, wherein the protein of interest is fused with a mutant of a human AGT, comprising:

(a) contacting the mutant of the human AGT fusion protein with a mixture of

(i) a compound of formula (1) wherein  $R_1$  additionally comprises  $R_5$ , wherein  $R_5$  is a substituted or unsubstituted cycloalkyl, cycloalkenyl or heterocyclyl group which does not react with the mutant AGT; and

(ii) another compound of formula (1), which reacts with the mutant AGT fusion protein; and

(b) detecting the mutant AGT fusion protein using the label in a system designed for recognizing and/or handling the label.